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(FILE 'HOME' ENTERED AT 19:19:41 ON 28 SEP 1998)

FILE 'ADISALERTS, ADISINSIGHT, AIDSLINE, BIOSIS, CANCERLIT, CAPLUS, CEN, DISSABS, DRUGLAUNCH, DRUGMONOG2, DRUGNL, EMBAL, EMBASE, IFIPAT, IPA, JICST-EPLUS, LIFESCI, MEDLINE, NAPRALERT, NLDB, PHIC, PHIN, PNI, SCISEARCH, TOXLINE, TOXLIT, USPATFULL' ENTERED AT 19:19:48 ON 28 SEP 1998

E BERNFIELD M/AU

L1 756 S E3-E9
 L2 1 S L1 AND MELANOCORTIN
 L3 1919 S MELANOCORTIN
 L4 64 S L3 AND (TRANSGEN? OR KNOCKOUT)
 L5 27 DUPLICATE REMOVE L4 (37 DUPLICATES REMOVED)
 L6 2054 S SYNDECAN
 L7 751 S L6 AND (MOUSE OR MICE)
 L8 43 S L7 AND (TRANSGEN? OR KNOCKOUT)
 L9 18 DUPLICATE REMOVE L8 (25 DUPLICATES REMOVED)
 E REIZES O/AU
 L10 38 S E3-E4
 L11 15 DUPLICATE REMOVE L10 (23 DUPLICATES REMOVED)
 L12 1413 S L3 AND RECEPTOR
 L13 179 S L12 AND (TARGET? OR DISRUPT?)
 L14 85 S L13 AND (MOUSE OR MICE)
 L15 54 S L14 AND OBES?
 L16 4 S L6 AND OBES?
 L17 234 S L12 AND OBES?
 L18 102 DUPLICATE REMOVE L17 (132 DUPLICATES REMOVED)
 L19 64 S L18 AND (MOUSE OR MICE)
 L20 5 S L19 NOT PY>1996
 L21 876 S L6 AND SYNDECAN-1
 L22 161 S L21 AND SEQUENCE
 L23 45 S L22 AND DNA
 L24 105 S L22 AND (DNA OR GENE)
 L25 66 S L24 AND (MOUSE OR MURINE OR MICE)

08/965,356

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(FILE 'HOME' ENTERED AT 10:18:45 ON 29 SEP 1998)

FILE 'ADISALERTS, ADISINSIGHT, AIDSLINE, BIOSIS, CANCERLIT, CAPLUS, CEN, DGENE, DISSABS, DRUGB, DRUGLAUNCH, DRUGMONOG2, DRUGNL, DRUGU, EMBAL, EMBASE, IFIPAT, IPA, JICST-EPLUS, KOSMET, LIFESCI, MEDLINE, NAPRALERT, NLDB, PHIC, PHIN, PNI, SCISEARCH, TOXLINE, ...' ENTERED AT 10:19:02 ON 29 SEP 1998

L1 130307 S (CYTOMEGALOVIRUS OR CMV)
L2 12576 S L1 AND PROMOTER
L3 3682 S L2 AND ENHANCER
L4 2585 S L3 AND (SEQUENC? OR CLON?)
L5 107203 S PROMOTER(10N)SEQUENC?
L6 1397 S L4 AND PROMOTER(10N)SEQUENC?
L7 587 S L6 AND CMV(10N)PROMOTER
L8 130 S L7 AND (CMV PROMOTER(10N)SEQUENC?)
L9 35 S L8 AND PROMOTER SEQUENCE
L10 8 S L3 AND CMV PROMOTER SEQUENCE
L11 2400 S L2 AND CMV PROMOTER
L12 42 S L11 AND CMV PROMOTER SEQUENCE
L13 42 DUPLICATE REMOVE L12 (0 DUPLICATES REMOVED)

L7 ANSWER 7 OF 587 BIOSIS COPYRIGHT 1998 BIOSIS
 ACCESSION NUMBER: 94:61784 BIOSIS
 DOCUMENT NUMBER: 97074784
 TITLE: Heterologous and homologous protection against
 influenza A by DNA vaccination: Optimization of
 DNA vectors.
 AUTHOR(S): Montgomery D L; Shiver J W; Leander K R; Perry H
 C; Friedman A; Martinez D; Ulmer J B; Donnelly J
 J; Liu M A
 CORPORATE SOURCE: Dep. Virus and Cell Biol., SumneytownPike
 WP16-101, Merck Res. Lab., West Point, PA 19486,
 USA
 SOURCE: DNA and Cell Biology 12 (9). 1993. 777-783. ISSN:
 1044-5498
 LANGUAGE: English

QH442.D2

AB We have recently shown that direct injection of DNA can be an effective vaccine strategy eliciting both humoral and cell-mediated immune responses. Vectors were designed specifically for vaccination by direct DNA injection and refined to improve plasmid production in *Escherichia coli*. The vectors consist of a pUC-19 backbone with the **cytomegalovirus (CMV) IE1 enhancer, promoter**, and intron A transcription regulatory elements and the BGH polyadenylation **sequences** driving the expression of the reporter gene CAT or influenza A nucleoprotein (NP) or hemagglutinin (HA). The respective vectors expressed high levels of chloramphenicol acetyltransferase (CAT) and NP in tissue culture, and yielded 14-15 mg of purified plasmid per liter of *Escherichia coli* culture. Immunization of mice with the NP and HA expression vectors resulted in protection from subsequent lethal challenges of influenza using either heterologous or homologous strains, respectively.

Nature 316 : 744
Dynan + Tjian

L7 ANSWER 10 OF 587 CANCERLIT

ACCESSION NUMBER: 1998285784 CANCERLIT

DOCUMENT NUMBER: 98285784

TITLE: Up to 100-fold increase of apparent gene expression
in the presence of Epstein-Barr virus oriP
sequences and EBNA1: implications of the
nuclear import of plasmids.

AUTHOR: Langle-Rouault F; Patzel V; Benavente A; Taillez M;
Silvestre N; Bompard A; Sczakiel G; Jacobs E; Rittner
K

CORPORATE SOURCE: Transgene S.A., 67000 Strasbourg, France.

SOURCE: JOURNAL OF VIROLOGY, (1998). Vol. 72, No. 7, pp.
6181-5.

Journal code: KCV. ISSN: 0022-538X.

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)

FILE SEGMENT: MEDL; Cancer Journals; L; Priority Journals

LANGUAGE: English

OTHER SOURCE: MEDLINE 98285784

ENTRY MONTH: 199808

AB A 100-fold increase in luciferase activity was observed in 293 cells, stably expressing Epstein-Barr nuclear antigen 1 (EBNA1; 293-EBNA1 cells), that had been transiently transfected with plasmids carrying Epstein-Barr virus (EBV) oriP **sequences**. This increase was observed in comparison to reporter gene activity obtained after transfection with a plasmid carrying no oriP **sequences**. The luciferase gene on these plasmids was under the control of either the **cytomegalovirus** immediate-early 1 gene **enhancer-promoter** (CMV IE1) or the Rous sarcoma virus **promoter**. The increase of reporter gene activity was not due to plasmid replication, since a similar enhancement was observed in the presence of aphidicolin, an inhibitor of replicative DNA synthesis, or after deletion of the dyad symmetry (DS) element within oriP. Luciferase production was not increased in the presence of only the DS element. Microinjection of plasmids carrying the **CMV IE1 promoter**-driven luciferase gene with or without oriP **sequences** into the nuclei of 293-EBNA1 cells resulted in a 17-fold increase in luciferase activity. Cytoplasmic injection of these plasmids led to an enhancement of luciferase activity of up to 100-fold. This difference in the factor of activation after nuclear or cytoplasmic injection could be ascribed to increased transport of plasmids carrying oriP from the cytoplasm to the nucleus in the presence of EBNA1. These data suggest the possibility of substantially increasing the apparent expression of a gene under the control of a strong constitutive **promoter** in the presence of oriP **sequences** and EBNA1. This improvement in expression is due to intranuclear enhancement of gene expression. oriP-specific transport of plasmid DNA from the cytoplasm of 293-EBNA1 cells to the nucleus seems to contribute to the observed effect.

QR355.J65

II. Useful **Promoter** Units for Reporter Genes

The reporter gene should be operatively associated with a **promoter** unit capable of being stimulated by a viral transacting transcription activator as described herein. Useful promoters include the human **cytomegalovirus** major intermediate-early **promoter** (hCMV-MIE) or the adenovirus early **promoter** (E1A, E1B **promoter**), or the adenovirus late region **promoter**. Preferably, the **CMV-MIE promoter** is a intron-free form of the **promoter**, so-called the **CMV-MIE "short" promoter**. **CMV promoter sequences** or plasmids containing them can be purchased commercially, e.g. from Invitrogen, Inc., Palo Alto (pCDM8) and from **Clontech**, Inc., Palo Alto. Preferably, the transcription further is stimulated by the inclusion of a cis-acting **enhancer sequence**, e.g., the mouse mammary tumor virus long terminal repeat (MMTV-LTR) or the Rous sarcoma virus long terminal repeat (RSV-LTR.) **Enhancer sequences** or plasmids containing them also are commercially available (e.g., from Invitrogen Inc., San Diego, or **Clontech** Inc., Palo Alto.) and/or also are available through the ATCC and ECACC.

III. Useful viral expression effector genes

The viral expression effector genes useful in the methods and cell lines of the invention are competent to act on the **promoter** that induces transcription of the reporter gene and/or to act on the reporter gene's transcript or the translation machinery.

At least one of the expression effector genes is a viral transacting transcription activator. Useful **sequences** include those encoded by the adenovirus -2E1A and E1B genes, as well as by the bovine papilloma virus early region DNA. Details on these **sequences** and vectors carrying these **sequences** can be found in Maat, J. et al. (1979) Gene 6:75, and in EP 0378,382 and Cockett, (1990) Nucleic Acids Research 19:319-325 all incorporated herein by reference. Whole bovine papilloma virus DNA can be obtained commercially, e.g., from IBI., New Haven (Catalog #3320.)

The authors of EP 0378,382 state that appropriate levels of the transcription activator can be obtained by choice of a suitable **promoter/enhancer** unit for its transcription (e.g., a weak **promoter** is preferred and a stable transcription activator expressing cell is produced before transfection with the reporter gene.) Alternatively, and as currently preferred herein, the activator gene is simultaneously co-transfected together with the reporter gene, and the transfected cells individually allowed to determine the appropriate, combined level of all recombinant, expressed genes, including the optimal level of the activator gene product for that cell when present in the cell in combination with the reporter gene and gene product.

L13 ANSWER 25 OF 42 DGENE COPYRIGHT 1998 DERWENT INFORMATION LTD
 ACCESSION NUMBER: 98N-V14004 cDNA DGENE
 TITLE: DNA containing inactive, mutated binding site for Gfi-1
 transcription repressor - used to increase gene
 expression in vitro or in vivo, e.g. in gene therapy
 INVENTOR: Grimes H L; Tsichlis P; Zweidler-Mckay P
 PATENT ASSIGNEE: (FOXC-N)FOX CHASE CANCER CENT
 PATENT INFO: WO 9748720 A1 971224 44 pp
 APPLICATION INFO: WO 97-US10486 970617
 PRIORITY INFO: US 96-19808 960617
 DOCUMENT TYPE: Patent
 LANGUAGE: English
 OTHER SOURCE: 98-063073 [06]

AB This is a mutant **cytomegalovirus (CMV)**
promoter sequence comprising mutated Gfi-1
 (growth factor independence-1) transcription repressor binding
 sites. This **promoter** is derived from the **CMV**
 -MIE wild-type **promoter** and is used in a novel isolated
 DNA construct which contains at least one mutated binding site for
 a Gfi-1 transcription repressor that hinders or prevents binding of
 Gfi-1 to this site. The expression vector contains an expression
 regulatory segment that contains at least one copy of the sequences
 shown in V19671 to V19685 linked operably to a coding segment
 selected from a group of cytokines, interleukins, interferons,
 growth factors and proto-oncogenes, and an isolated DNA molecule
 containing one of two 500 bp sequences shown in V14003 and V14004.
 Altering the binding site increases expression of these genes
 controlled by regulators that include binding sites for Gfi-1 both
 in cultured cells and in vivo (for gene therapy or DNA
 vaccination). A vector containing a normal gene under control of a
 regulator with the mutated binding site can be administered to a
 patient having a disease associated with an aberrant form of the
 gene

L5 ANSWER 24 OF 27 MEDLINE

ACCESSION NUMBER: 97148695 MEDLINE

DOCUMENT NUMBER: 97148695

TITLE: Targeted disruption of the **melanocortin-4** receptor results in obesity in mice.

AUTHOR: Huszar D; Lynch C A; Fairchild-Huntress V; Dunmore J H; Fang Q; Berkemeier L R; Gu W; Kesterson R A; Boston B A; Cone R D; Smith F J; Campfield L A; Burn P; Lee F

CORPORATE SOURCE: Millennium Pharmaceuticals, Inc., Cambridge, Massachusetts 02139, USA.

SOURCE: CELL, (1997 Jan 10) 88 (1) 131-41.
Journal code: CQ4. ISSN: 0092-8674.

PUB. COUNTRY: United States

Journal; Article; (JOURNAL ARTICLE)

LANGUAGE: English

FILE SEGMENT: Priority Journals; Cancer Journals

ENTRY MONTH: 199704

ENTRY WEEK: 19970403

AB The **melanocortin-4** receptor (MC4-R) is a G protein-coupled, seven-transmembrane receptor expressed in the brain. Inactivation of this receptor by gene targeting results in mice that develop a maturity onset obesity syndrome associated with hyperphagia, hyperinsulinemia, and hyperglycemia. This syndrome recapitulates several of the characteristic features of the agouti obesity syndrome, which results from ectopic expression of agouti protein, a pigmentation factor normally expressed in the skin. Our data identify a novel signaling pathway in the mouse for body weight regulation and support a model in which the primary mechanism by which agouti induces obesity is chronic antagonism of the MC4-R.

L5 ANSWER 22 OF 27 CAPLUS COPYRIGHT 1998 ACS
ACCESSION NUMBER: 1997:718678 CAPLUS
DOCUMENT NUMBER: 128:21146
TITLE: Obesity and the adipocyte. Role of the agouti
gene in obesity
AUTHOR(S): Michaud, E. J.; Mynatt, R. L.; Miltenberger, R.
J.; Klebig, M. L.; Wilkinson, J. E.; Zemel, M.
B.; Wilkison, W. O.; Woychik, R. P.
CORPORATE SOURCE: Life Sci. Div., Oak Ridge Natl. Lab., Oak Ridge,
TN, 37831, USA
SOURCE: J. Endocrinol. (1997), 155(2), 207-209
CODEN: JOENAK; ISSN: 0022-0795
PUBLISHER: Journal of Endocrinology
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English

AB A review, with .apprx.40 refs., suggesting that the wild-type agouti protein acts on both the CNS and tissues in the periphery to induce the obesity syndrome. In the CNS agouti may antagonize neural **melanocortin** receptor(s), resulting in obesity, hyperphagia, and hyperinsulinemia, as obsd. in MC4-R **knockout** mice. In the periphery, agouti expression in adipose tissue, coupled with insulin treatment, results in significant wt. gains in mice. Given that hyperphagia appears to be an important aspect of the agouti-induced obesity syndrome, it is noteworthy that pancreatic beta-cell hyperplasia precedes obesity in mutant agouti mice. In addn., increases in $[Ca^{2+}]_i$ in beta cells stimulate insulin release. Therefore, it is possible that ectopic expression of the agouti gene in the pancreas may act directly on the beta cells to trigger hyperplasia.

L15 ANSWER 10 OF 54 CAPLUS COPYRIGHT 1998 ACS
 ACCESSION NUMBER: 1997:509759 CAPLUS
 DOCUMENT NUMBER: 127:203857
 TITLE: Molecular screening of the human
melanocortin-4 receptor gene.
 Identification of a missense variant showing no
 association with **obesity**, plasma
 glucose, or insulin
 AUTHOR(S): Gotoda, T.; Scott, J.; Aitman, T. J.
 CORPORATE SOURCE: Royal Postgraduate Medical School, Hammersmith
 Hospital, London, W12 0NN, UK
 SOURCE: Diabetologia (1997), 40(8), 976-979
 CODEN: DBTG AJ; ISSN: 0012-186X
 PUBLISHER: Springer
 DOCUMENT TYPE: Journal
 LANGUAGE: English
 AB **Disruption** of the **melanocortin-4** (MC-4)
receptor gene in **mice** results in maturity-onset
obesity, hyperinsulinemia and hyperglycemia. These
 phenotypes are characteristic of human **obesity** that
 frequently accompanies non-insulin-dependent diabetes. It is
 therefore possible that human MC-4 **receptor** gene mutations
 contribute to human **obesity**. To test this possibility, we
 examd. by DNA sequencing the entire coding region of the human MC-4
receptor gene in 40 morbidly **obese** (BMI >35 kg/m²)
 white British males and examd. the 5'- and 3'flanking regions in 20
 out of these **obese** subjects. We also sequenced all these
 regions in 10 lean (BMI <18 kg/m²) white British males for a ref.
 We identified a single nucleotide substitution that replaces valine
 with isoleucine at codon 103, in two **obese** subjects in the
 heterozygous state. No other nucleotide alterations were found.
 The prevalence of this missense variant was studied in 322 white
 British males (190 with BMI >28 kg/m² and 132 with BMI < 22kg/m²)
 selected from a population-based epidemiol. survey. In these
 subjects, no homozygotes for the isoleucine allele were found. The
 frequency of heterozygotes was similar (4.2 vs. 4.5%) in the two
 groups and there was no significant difference in BMI, total
 skinfold thickness, plasma insulin and glucose levels between
 heterozygotes and codon-103 valine homozygotes in either group.
 These results suggest that coding sequence mutations in the MC-4
receptor gene are unlikely to be a major cause of human
obesity, at least in white British males [Diabetologia
 (1997) 40: 976-979].

L15 ANSWER 8 OF 54 CAPLUS COPYRIGHT 1998 ACS

ACCESSION NUMBER: 1998:404594 CAPLUS

DOCUMENT NUMBER: 129:170602

TITLE: Recently identified peptides involved in the regulation of body weight

AUTHOR(S): Bessesen, Daniel H.; Faggioni, Raffaella

CORPORATE SOURCE: University of Colorado Health Sciences Center and Denver Health Medical Center, Denver, CO, 80204-4507, USA

SOURCE: Semin. Oncol. (1998), 25(2, Suppl. 6), 28-32
CODEN: SOLGAV; ISSN: 0093-7754

PUBLISHER: W. B. Saunders Co.

DOCUMENT TYPE: Journal; General Review

LANGUAGE: English

AB A review, with 27 refs. The application of mol. and genetic techniques to the study of body wt. regulation have produced exciting new insights into the physiol. systems governing energy expenditure, appetite, and metabolic signaling. A no. of new peptides have been identified that play important roles in these regulatory systems. These include the hormone leptin, the short and long forms of the leptin **receptor**; uncoupling proteins, agouti protein, **melanocortin receptor** isoforms, melanin-concg. hormone, and the proteins responsible for tub and fat, two monogenic **mouse** models of **obesity**. This article reviews some of the new insights gained from studies of these peptides. Although much of this new knowledge has come from studies of **obesity**, there may be implications for the clin. syndromes assocd. with wt. loss. As more is learned about these systems, potential new **targets** for therapeutic intervention will likely become evident. These interventions may develop first as **obesity** treatments, but investigators and clinicians involved in the care of cachectic patients should follow these scientific developments as well.

L19 ANSWER 2 OF 64 BIOSIS COPYRIGHT 1998 BIOSIS

ACCESSION NUMBER: 98:386925 BIOSIS

DOCUMENT NUMBER: 01386925

TITLE: Evidence that orexigenic effects of

melanocortin 4 receptor

antagonist HS014 are mediated by neuropeptide Y.

AUTHOR(S): Kask A; Rago L; Korrovits P; Wikberg J E S;
Schioth H B

CORPORATE SOURCE: Dep. Pharmacol., Univ. Tartu Ulikooli 18, Tartu
EE-2400, Estonia

SOURCE: Biochemical and Biophysical Research
Communications 248 (2). 1998. 245-249. ISSN:
0006-291X

LANGUAGE: English

AB Recent studies using **melanocortin-4 receptor**

(MC4R) knockout **mice** and MC4R antagonists have shown that weakening of MC4R-ergic tone increases food intake and causes **obesity**. In this study, we used the newly discovered selective MC4R antagonist HS014 for increasing food intake in free-feeding rats and evaluated the effects of the NPY Y-1 **receptor** antagonist 1229U91 and the selective serotonin uptake inhibitor fluoxetine on this increased feeding behavior. 1229U91 (12 nmol, i.c.v.), which alone does not affect food intake, significantly attenuated the orexigenic effects of HS014, whereas 1 and 3 nmol doses of 1229U91 were ineffective. Fluoxetine, which has been shown to inhibit NPY release, inhibited spontaneous food intake and completely blocked the stimulation of food intake by HS014. These data suggest that feeding induced by weakening of the MC4R-ergic tone may be mediated through activation of the NPY-ergic system. This is the first report showing that physiological feeding response evoked by MC4R blockage is influenced by NPY signalling.

L19 ANSWER 6 OF 64 BIOSIS COPYRIGHT 1998 BIOSIS
ACCESSION NUMBER: 97:505006 BIOSIS
DOCUMENT NUMBER: 99804209
TITLE: Antagonism of central **melanocortin**
receptors in vitro and in vivo by Agouti-related
protein.
AUTHOR(S): Ollmann M M; Wilson B D; Yang Y-K; Kerns J A; Chen
Y; Gantz I; Barsh G S
CORPORATE SOURCE: Beckman Cent. B271A, Stanford Univ. Sch. Med.,
Stanford, CA 94305-5323, USA
SOURCE: Science (Washington D C) 278 (5335). 1997.
135-138. ISSN: 0036-8075
LANGUAGE: English
AB Expression of Agouti protein is normally limited to the skin where it
affects pigmentation, but ubiquitous expression causes
obesity. An expressed sequence tag was identified that
encodes Agouti-related protein, whose RNA is normally expressed in
the hypothalamus and whose levels were increased eightfold in ob/ob
mice. Recombinant Agouti-related protein was a potent,
selective antagonist of Mc3r and Mc4r, **melanocortin**
receptor subtypes implicated in weight regulation. Ubiquitous
expression of human AGRP complementary DNA in transgenic **mice**
caused **obesity** without altering pigmentation. Thus,
Agouti-related protein is a neuropeptide implicated in the normal
control of body weight downstream of leptin signaling.

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L19 ANSWER 7 OF 64 BIOSIS COPYRIGHT 1998 BIOSIS

ACCESSION NUMBER: 97:487558 BIOSIS

DOCUMENT NUMBER: 99786761

TITLE: Genetics of human **obesity**: Research directions.

AUTHOR(S): Bray G; Bouchard C

CORPORATE SOURCE: Pennington Biomed. Res. Cent., Baton Rouge, LA 70808-4124, USA

SOURCE: FASEB Journal 11 (12). 1997. 937-945. ISSN: 0892-6638

LANGUAGE: English

AB Rapid strides in understanding the physiology controlling energy or nutrient intake and energy expenditure have complemented the search for the genetic basis of **obesity**. Several single gene defects are known that produce **obesity** in animals. All of these have been cloned within the past 4 years, providing a rich new base for understanding **obesity**. Since **obesity** is likely to be "multifactorial," a number of laboratories have used the quantitative trait locus (QTL) technique of genome scanning to identify candidate genomic regions and, eventually, genes that may influence body weight and body fat. So far, 18 QTLs have been identified in association with crossbreeding strains of **mice** or rats with variable susceptibility to **obesity**. A number of mendelian disorders are known to exist in humans, but no specific genes have yet been identified for them. The potential for inserting new genetic material into mammals has produced numerous transgenic **mice** with increased or decreased quantities of body fat.

These models will provide a continuing source of new insights into **obesity**. Several areas in the human genome have been linked to the development of **obesity**. Among the candidate genes with evidence of linkage to body fat are TNF-alpha, adenosine deaminase, and **melanocortin-3 receptor**. The new insights described above have invigorated the pharmaceutical industry to increase their efforts for new drug development aimed at the growing problem of **obesity**.

L19 ANSWER 9 OF 64 BIOSIS COPYRIGHT 1998 BIOSIS
 ACCESSION NUMBER: 97:453920 BIOSIS
 DOCUMENT NUMBER: 99753123
 TITLE: Homologous pigmentation mutations in human,
mouse and other model organisms.
 AUTHOR(S): Jackson I J
 CORPORATE SOURCE: MRC Human Genetics Unit, Western General Hosp.,
 Crewe Road, Edinburgh EH4 2XU, UK
 SOURCE: Human Molecular Genetics 6 (10). 1997. 1613-1624.
 ISSN: 0964-6906
 LANGUAGE: English

AB **Mouse** coat colour genes have long been studied as a paradigm for genetic interactions in development.. A number of these genes have been cloned and most correspond to human genetic disease loci. The proteins encoded by these genes include transcription factors, **receptor** tyrosine kinases and growth factors, G-protein coupled receptors and their ligands, membrane proteins, structural proteins and enzymes. Many of the mutations have pleiotropic effects, indicating that these proteins play a wider role in developmental or cellular processes. In this review I tabulate the available data on all pigmentation genes cloned from **mouse** or human, and I focus on three particular systems. One family of genes, including LYST and HPS/ep, shows the relationship between melanosomes and lysosomes. The G-protein coupled **receptor**, endothelin **receptor**-B, and its ligand, endothelin-3, are required for the development of both melanocytes and enteric neurons. The **melanocortin-1 receptor** is expressed only on melanocytes, but mutations that cause overexpression of agouti protein, an antagonist of the **receptor**, result in **obesity**, and highlight a role of melanocortins in weight homoeostasis.

L19 ANSWER 17 OF 64 BIOSIS COPYRIGHT 1998 BIOSIS
ACCESSION NUMBER: 97:179881 BIOSIS
DOCUMENT NUMBER: 99471594
TITLE: Mutations in the carboxyl terminus of the agouti protein decrease agouti inhibition of ligand binding to the **melanocortin** receptors.
AUTHOR(S): Kiefer L L; Ittoop O R R; Bunce K; Truesdale A T; Willard D H; Nichols J S; Blanchard S G; Mountjoy K; Chen W-J; Wilkison W O
CORPORATE SOURCE: Glaxo Wellcome, 5 Moore Dr., V211, Research Triangle Park, NC 27709, USA
SOURCE: Biochemistry 36 (8). 1997. 2084-2090. ISSN: 0006-2960
LANGUAGE: English
AB Several mutations that cause ectopic expression of the agouti gene result in **obesity**, hyperinsulinemia, and yellow coat color. A candidate pathway for agouti induced **obesity** and hyperinsulinemia is through altered signaling by **melanocortin** receptors, as agouti normally regulates coat coloration through antagonism of **melanocortin receptor 1**. Furthermore, **melanocortin** peptides mediate functions including steroidogenesis, lipolysis, and thermoregulation. We report apparent inhibition dissociation constants for **mouse** and human agouti protein inhibition of ligand binding to the **melanocortin** receptors, to determine which of these receptors might be involved in agouti induced diabetes. The similarity in the apparent K-I values for agouti inhibition of ligand binding to the brain **melanocortin** receptors 3 and 4 (**mouse**: K-I app = 190 +- 74 and 54 +- 18 nM; human: K-I app = 140 +- 56 and 70 +- 18 nM, respectively) suggests that the MC3-R is a potential candidate for a **receptor** mediating the effects of agouti protein overexpression. Agouti residues important for **melanocortin receptor** inhibition were identified through the analysis of deletion constructs and site-specific variants. Val83 is important for inhibition of binding to MC1-R (K-I app for Val83Ala agouti increased 13-fold relative to wild-type protein). Arg85, Pro86, and Pro89 are important for selective inhibition of binding between MC1-R and MC3-R and MC4-R as their apparent K-I values are essentially unchanged at MC1-R, while they have increased 6-10-fold relative to wild-type protein at MC3-R and MC4-R.

L19 ANSWER 20 OF 64 BIOSIS COPYRIGHT 1998 BIOSIS
ACCESSION NUMBER: 96:461724 BIOSIS
DOCUMENT NUMBER: 99184080
TITLE: Coupled site-directed mutagenesis-transgenesis
identifies important functional domains of the
mouse agouti protein.
AUTHOR(S): Perry W L; Nakamura T; Swing D A; Secrest L;
Eagleson B; Hustad C M; Copeland N G; Jenkins N A
CORPORATE SOURCE: Mammalian Genetics Lab., ABL-Basic Research
Program, NCI-Frederick Cancer Research Development
Center, P.O. Box B, Frederick, MD 21702, USA
SOURCE: Genetics 144 (1). 1996. 255-264. ISSN: 0016-6731
LANGUAGE: English
AB The agouti locus encodes a novel paracrine signaling molecule
containing a signal sequence, an N-linked glycosylation site, a
central lysine-rich basic domain, and a C-terminal tail containing 10
cysteine (Cys) residues capable of forming five disulfide bonds. When
overexpressed, agouti causes a number of pleiotropic effects
including yellow coat and adult-onset **obesity**. Numerous
studies suggest that agouti causes yellow coat color by antagonizing
the binding of alpha-melanocyte-stimulating hormone (alpha-MSH) to
the alpha-MSH-(**melanocortin-1**) **receptor**. With the
goal of identifying functional domains of agouti important for its
diverse biological activities, we have generated 14 agouti mutations
by in vitro site-directed mutagenesis and analyzed these mutations in
transgenic **mice** for their effects on coat color and
obesity. These studies demonstrate that the signal sequence,
the N-linked glycosylation site, and the C-terminal Cys residues are
important for full biological activity, while at least a portion of
the lysine-rich basic domain is dispensable for normal function. They
also show that the same functional domains of agouti important in
coat color determination are important for inducing **obesity**
, consistent with the hypothesis that agouti induces **obesity**
by antagonizing **melanocortin** binding to other
melanocortin receptors.

L20 ANSWER 4 OF 5 BIOSIS COPYRIGHT 1998 BIOSIS

ACCESSION NUMBER: 94:546689 BIOSIS

DOCUMENT NUMBER: 98006237

TITLE: Agouti protein is an antagonist of the melanocyte-stimulating-hormone **receptor**.

AUTHOR(S): Lu D; Willard D; Patel I R; Kadwell S; Overton L; Kost T; Luther M; Chen W; Woychik R P; Wilkison W O; Cone R D

CORPORATE SOURCE: Div. Mol. Sci., Glaxo Res. Inst., Research Triangle Park, NC 27709, USA

SOURCE: Nature (London) 371 (6500). 1994. 799-802. ISSN: 0028-0836

LANGUAGE: English

AB The genetic loci agouti and extension control the relative amounts of eumelanin (brown-black) and phaeomelanin (yellow-red) pigments in mammals: extension encodes the **receptor** for melanocyte-stimulating hormone (MSH) and agouti encodes a novel 131-amino-acid protein containing a signal sequence-3,4. Agouti, which is produced in the hair follicles, acts on follicular melanocytes-6 to inhibit alpha-MSH-induced eumelanin production, resulting in the subterminal band of phaeomelanin often visible in mammalian fur. Here we use partially purified agouti protein to demonstrate that agouti is a high-affinity antagonist of the MSH **receptor** and blocks alpha-MSH stimulation of adenylyl cyclase, the effector through which alpha-MSH induces eumelanin synthesis. Agouti was also found to be an antagonist of the **melanocortin-4 receptor**, a related MSH-binding **receptor**. Consequently, the **obesity** caused by ectopic expression of agouti in the lethal yellow (Al) **mouse** may be due to the inhibition of **melanocortin receptor(s)** outside the hair follicle.

L25 ANSWER 4 OF 66 BIOSIS COPYRIGHT 1998 BIOSIS
 ACCESSION NUMBER: 93:432578 BIOSIS
 DOCUMENT NUMBER: BA96:87203
 TITLE: STRUCTURAL ORGANIZATION AND GENOMIC

**SEQUENCE OF MOUSE
 SYNDECAN-1 GENE.**

AUTHOR(S): VIHINEN T; AUVINEN P; ALANEN-KURKI L; JALKANEN M
 CORPORATE SOURCE: TURKU CENTER BIOTECHNOL., TYKISTOEKATU 6, BIO CITY,
 P.O. BOX 123, SF-20521 TURKU, FINL.
 SOURCE: J BIOL CHEM 268 (23). 1993. 17261-17269. CODEN:
 JBCHA3 ISSN: 0021-9258
 LANGUAGE: English

AB **Syndecan-1** is an integral membrane proteoglycan, which binds several extracellular matrix components and growth factors. Its expression follows morphogenetic rather than histological patterns during embryonic development and is regulated by epithelial-mesenchymal interactions during organogenesis. Malignant transformation has been shown to suppress **syndecan-1** expression. In order to understand better the regulation of **syndecan-1** expression, we have determined the structural organization of **mouse syndecan-1 gene**. Several genomic clones were isolated, covering the entire 23-kilobase (kb) **syndecan-1 gene**. All five exons, four introns, and the 5'- and 3'-flanking regions were sequenced. The first intron was very long (17,582 base pairs (bp)) if compared with the others that were only a few hundred nucleotides in length. The first exon contained only the signal **sequence** and exons II-IV all the glycosaminoglycan binding sites. The fifth exon resided both transmembrane and cytoplasmic domains, which are known to be conserved among the members of the **syndecan** family. This genomic structure explains why these members could have heterologous extracellular domains and homologous transmembrane and cytoplasmic domains. **Syndecan-1 gene** was shown by primer extension analysis to have three transcription initiation sites which were confirmed by polymerase chain reaction. These initiation sites were found to locate -217, -266, and -591 bp from described cDNA (Saunders, S., Jalkanen, M., O'Farrell, S., and Bernfield, M. (1989) J. Cell Biol. 108, 1547-1556). Within the 5'-end of the **gene** a 2000-bp-long CpG nucleotide-rich **sequence** resembling a CpG island was found, which started from the transcription initiation sites and ended in the first intron. At the 3'-end of the **gene** an other polyadenylation signal **sequence** was revealed 638 bp downstream from the first one. The two mRNAs (2.6 kb and 3.4 kb) were shown to be produced by alternative polyadenylation.

L5 ANSWER 1 OF 27 CAPLUS COPYRIGHT 1998 ACS

ACCESSION NUMBER: 1998:324878 CAPLUS

DOCUMENT NUMBER: 128:318018

TITLE: Construction of **transgenic** mice
expressing a syndecan 1 gene in regions of
hypothalamus for wt. regulation and therapy

INVENTOR(S): Bernfield, Merton; Reizes, Ofer

PATENT ASSIGNEE(S): Children's Medical Center Corp., USA

SOURCE: PCT Int. Appl., 61 pp.

CODEN: PIXXD2

	NUMBER	DATE
	-----	-----
PATENT INFORMATION:	WO 9820121 A1	19980514
DESIGNATED STATES:	W: AL, AU, BA, BB, BG, BR, CA, CN, CU, CZ, EE, GE, HU, IL, IS, JP, KP, KR, LC, LK, LR, LT, LV, MG, MK, MN, MW, MX, NO, NZ, PL, RO, SG, SI, SK, SL, TR, TT, UA, UZ, VN, YU, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FI, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG	
APPLICATION INFORMATION:	WO 97-US20003	19971106
PRIORITY APPLN. INFO.:	US 96-30758	19961106
DOCUMENT TYPE:	Patent	
LANGUAGE:	English	

AB Lines of **transgenic** mice have been developed which preferentially express a syndecan in the regions of the hypothalamus which are known to be important in wt. control. The animals were made using a construct including a cytomegalovirus promoter and the 3' untranslated region, including the polyadenylation site, of the bovine growth hormone gene, as well as cDNA encoding syndecan-1. The mice express the syndecan-1 **transgene** in many tissues, with the expression in the brain occurring preferentially in their hypothalamus. The mice are characterized by elevated levels of circulating syndecan-1 ectodomain and exhibit enormous wt. gain after reaching sexual maturity. **Transgenic** animals in which stop codons have been inserted into the construct so that the syndecan is not expressed do not exhibit the same enormous wt. gain. The animals have a relatively normal distribution of fat, are completely healthy and heterozygotes reproduce, and show other indicators assocd. with obesity in humans. The mice are useful in understanding the factors involved in wt. regulation and in designing and screening for drugs which are involved in wt. regulation and that can either enhance or reduce appetite and activity. Wasting disorders which can be examd. in these mice include idiopathic obesity, anorexia nervosa, and cachexia due to cancer, cancer chemotherapy, chronic inflammatory disease, rheumatoid and collagen diseases, and chronic infections.

L5 ANSWER 2 OF 27 USPATFULL

ACCESSION NUMBER: 1998:91815 USPATFULL

TITLE: Yeast cells engineered to produce pheromone
system protein surrogates, and uses therefor

INVENTOR(S): Fowlkes, Dana M., Chapel Hill, NC, United States

PATENT ASSIGNEE(S):

Broach, Jim, Princeton, NJ, United States
 Manfredi, John, Ossining, NY, United States
 Klein, Christine, Ossining, NY, United States
 Murphy, Andrew J., Montclair, NJ, United States
 Paul, Jeremy, South Nyack, NY, United States
 Trueheart, Joshua, South Nyack, NY, United States
 Cadus Pharmaceutical Corporation, Tarrytown, NY,
 United States (U.S. corporation)

	NUMBER	DATE
PATENT INFORMATION:	US 5789184	980804
APPLICATION INFO.:	US 95-464531	950605 (8)
RELATED APPLN. INFO.:	Continuation-in-part of Ser. No. US 94-322137, filed on 13 Oct 1994 which is a continuation-in-part of Ser. No. US 94-309313, filed on 20 Sep 1994, now abandoned which is a continuation-in-part of Ser. No. US 94-190328, filed on 31 Jan 1994, now abandoned which is a continuation-in-part of Ser. No. US 93-41431, filed on 31 Mar 1993, now abandoned	
DOCUMENT TYPE:	Utility	
PRIMARY EXAMINER:	Ketter, James	
ASSISTANT EXAMINER:	Yucel, Irem	
LEGAL REPRESENTATIVE:	Lahive & Cockfield, LLP; DeConti, Jr., Giulio A.; Kara, Catherine J.	
NUMBER OF CLAIMS:	48	
EXEMPLARY CLAIM:	1	
NUMBER OF DRAWINGS:	14 Drawing Figure(s); 13 Drawing Page(s)	
LINE COUNT:	6573	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

AB Yeast cells are engineered to express both a surrogate of a pheromone system protein (e.g., enzymes involved in maturation of .alpha.-factor, transporters of a-factor, pheromone receptors, etc.) and a potential peptide modulator of the surrogate, in such a manner that the inhibition or activation of the surrogate affects a screenable or selectable trait of the yeast cells. Various additional features improve the signal-to-noise ratio of the screening/selection system.

L5 ANSWER 3 OF 27 USPATFULL

ACCESSION NUMBER: 1998:68802 USPATFULL
 TITLE: Genes encoding art, an agouti-related transcript
 INVENTOR(S): Stark, Kevin Lee, Newbury Park, CA, United States
 Luethy, Roland, Newbury Park, CA, United States
 PATENT ASSIGNEE(S): Amgen Inc., Thousand Oaks, CA, United States
 (U.S. corporation)

	NUMBER	DATE
PATENT INFORMATION:	US 5766877	980616
APPLICATION INFO.:	US 96-757541	961127 (8)
DOCUMENT TYPE:	Utility	
PRIMARY EXAMINER:	Ulm, John	
ASSISTANT EXAMINER:	Teng, Sally P.	
LEGAL REPRESENTATIVE:	Oleski, Nancy A.; Levy, Ron K.; Odre, Steven M.	
NUMBER OF CLAIMS:	26	
EXEMPLARY CLAIM:	1	
NUMBER OF DRAWINGS:	11 Drawing Figure(s); 11 Drawing Page(s)	
LINE COUNT:	1681	

CAS INDEXING IS AVAILABLE FOR THIS PATENT.

AB Disclosed is a novel gene termed ART which is expressed primarily in selected regions of the brain, as well as adrenal and lung tissues. Polypeptides encoded by ART are also disclosed, as are methods for preparing ART DNA and amino acid sequences.

L5 ANSWER 4 OF 27 EMBAL COPYRIGHT 1998 ELSEVIER SCI. B.V.DUPLICATE 1
 ACCESSION NUMBER: 1998284749 EMBASE Alert (EMBAL)
 TITLE: Authentic cell-specific and developmentally regulated
 expression of pro- opiomelanocortin genomic fragments
 in hypothalamic and hindbrain neurons of
transgenic mice.
 AUTHOR: Young J.I.; Otero V.; Cerdan M.G.; Falzone T.L.; Chan
 E.C.; Low M.J.; Rubinstein M.
 CORPORATE SOURCE: Dr. M. Rubinstein, Inst. Invest. Ing. Gen. Biol.
 Molec., Consejo Nac. Invest. Cie./Tecnica, Vuelta de
 Obligado 2490, 1428 Buenos Aires, Argentina
 SOURCE: Journal of Neuroscience, (1 Sep 1998) 18/17
 (6631-6640). Refs: 36.
 CODEN: JNRSD ISSN: 0270-6474
 PUB. COUNTRY: United States
 DOCUMENT TYPE: Article
 LANGUAGE: English
 SUMMARY LANGUAGE: English

AB The pro-opiomelanocortin (POMC) gene is expressed in a subset of
 hypothalamic and hindbrain neurons and in pituitary melanotrophs and
 corticotrophs. POMC neurons release the potent opioid beta-endorphin
 and several active melanocortins that control homeostasis and
 feeding behavior. POMC gene expression in the CNS is believed to be
 controlled by distinct cis- acting regulatory sequences. To analyze
 the transcriptional regulation of POMC in neuronal and endocrine
 cells, we produced **transgenic** mice carrying POMC27*, a
transgene containing the entire 6 kb of the POMC
 transcriptional unit together with 13 kb of 5' flanking regions and
 8 kb of 3' flanking regions: POMC27* was tagged with a heterologous
 30 bp oligonucleotide in the third exon. In situ hybridization
 studies showed an accurate cell-specific pattern of expression of
 POMC27* in the arcuate nucleus and the pituitary. Hypothalamic
 mRNA-positive neurons colocalized entirely with beta-endorphin
 immunoreactivity. No ectopic **transgenic** expression was
 detected in the brain. Deletional analyses demonstrated that
 neuron-specific expression of POMC **transgenes** required
 distal 5' sequences localized upstream of the pituitary- responsive
 proximal cis-acting elements that were identified previously.
 POMC27* exhibited a spatial and temporal pattern of expression
 throughout development that exactly paralleled endogenous POMC.
 RNase protection assays revealed that POMC27* expression mimicked
 that of POMC in different areas of the CNS and most peripheral
 organs with no detectable ectopic expression. Hormonal regulation of
 POMC27* and POMC was identical in the hypothalamus and pituitary.
 These results show that distal 5' sequences of the POMC gene located
 between -13 and -2 kb target expression into the CNS of
transgenic mice in a precise neuron-specific,
 developmentally and hormonally regulated manner.

L5 ANSWER 5 OF 27 SCISEARCH COPYRIGHT 1998 ISI (R)
 ACCESSION NUMBER: 1998:380016 SCISEARCH
 THE GENUINE ARTICLE: ZN166
 TITLE: **Melanocortin** receptors and delta-opioid
 receptor mediate opposite signalling actions of
 POMC-derived peptides in CATH.a cells
 AUTHOR: Rene F; Muller A; Jover E; Kieffer B; Koch B;
 Loeffler J P (Reprint)
 CORPORATE SOURCE: UMR CNRS 7519, LAB NEUROPHYSIOL CELLULAIRE &
 INTEGREE, 21 RUE RENE DESCARTES, F-67084 STRASBOURG,
 FRANCE (Reprint); UMR CNRS 7519, LAB NEUROPHYSIOL
 CELLULAIRE & INTEGREE, F-67084 STRASBOURG, FRANCE;
 HOSPICES CIVIL STRASBOURG, CLIN DOULEUR, F-67000
 STRASBOURG, FRANCE; ECOLE SUPER BIOTECHNOL, UPR CNRS
 9050, LAB PROT & RECEPTEURS MEMBRANAIRES, F-67400

ILLKIRCH GRAFFENS, FRANCE
COUNTRY OF AUTHOR: FRANCE
SOURCE: EUROPEAN JOURNAL OF NEUROSCIENCE, (MAY 1998) Vol.
10, No. 5, pp. 1885-1894.
Publisher: OXFORD UNIV PRESS, GREAT CLARENDON ST,
OXFORD OX2 6DP, ENGLAND.
ISSN: 0953-816X.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: English
REFERENCE COUNT: 53

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB The locus coeruleus is innervated by proopiomelanocortin (POMC)-derived peptide immunoreactive fibres. The biological effects of alpha melanocyte-stimulating hormone (alpha MSH) and beta-endorphin on second messengers (cAMP, inositol phosphates) and gene transcription were studied in the locus coeruleus-derived cell line CATH.a.

RT-PCR analysis revealed the presence of four MSH receptor subtypes (1, 3, 4 and 5). Activation of these receptors by diacetyl alpha MSH stimulated cAMP accumulation in a dose-dependent manner (EC50: 4×10^{-9} M). Diacetyl alpha MSH stimulated transcription from reporter genes driven by the c-fos or tyrosine hydroxylase promoter. This effect was abolished when protein kinase A was inactivated with a dominant inhibitory mutant. RT-PCR analyses revealed the presence of delta-, but not mu- and kappa-opioid receptor. Pharmacological analysis showed that beta-endorphin (EC50: 2.5×10^{-8} M), but not N-acetyl beta-endorphin, antagonized the biological effect of diacetyl alpha MSH on cAMP production and gene transcription.

Since N-acetylation regulates the biological activity of alpha MSH and beta-endorphin in an opposite manner, we propose a model where the rate of secretion dictated by the bioelectric activity of the presynaptic neuron modulates POMC-derived peptide maturation and the resulting biological signal sensed by the postsynaptic plate.

L5 ANSWER 6 OF 27 SCISEARCH COPYRIGHT 1998 ISI (R)
ACCESSION NUMBER: 1998:654138 SCISEARCH
THE GENUINE ARTICLE: 112YB
TITLE: The role of the Agouti protein in the yellow mouse obesity syndrome
AUTHOR: Moussa N M (Reprint)
CORPORATE SOURCE: UNIV TENNESSEE, DEPT NUTR, 1215 W CUMBERLAND AVE, KNOXVILLE, TN 37996 (Reprint)
COUNTRY OF AUTHOR: USA
SOURCE: M S-MEDECINE SCIENCES, (AUG-SEP 1998) Vol. 14, No. 8-9, pp. 898-906.
Publisher: MASSON EDITEUR, 120 BLVD SAINT-GERMAIN, 75280 PARIS 06, FRANCE.
ISSN: 0767-0974.
DOCUMENT TYPE: Article; Journal
FILE SEGMENT: LIFE
LANGUAGE: French
REFERENCE COUNT: 46

ABSTRACT IS AVAILABLE IN THE ALL AND IALL FORMATS

AB Several dominant mutations at the agouti locus in the mouse cause a syndrome of marked obesity, hyperinsulinemia, insulin resistance, hyperglycemia, increased lean body mass as well as yellow coat color. Dominant obese yellow mutants, such as viable yellow (A(vy)) and let hal yellow (A(y)) exhibit mutations in the promoter region of the agouti gene, resulting in ectopic overexpression of agouti transcripts which contain the entire protein-coding portion in numerous tissues. **Transgenic** mice in which the wild-type agouti cDNA is placed under the transcriptional control of ubiquitous promoters develop the yellow mouse obesity syndrome,

demonstrating that ectopic expression of agouti per se is responsible for this syndrome. While expression of agouti in adipose tissue does not lead to obesity, the combination of hyperinsulinemia and agouti expression in adipose tissue leads to weight gain. The mechanism of agouti regulation of mouse coat color is based upon competitive antagonism of melanocyte stimulating hormone (alpha-MSH) binding at the **melanocortin** receptor 1 (MC1-R), resulting in suppression of cAMP production and a shift from eumelanin (black pigment) to pheomelanin (yellow pigment) production. Similar to its action on the skin, agouti acts centrally as an antagonist to alpha-MSH at the **melanocortin** receptor (MC-4R). Agouti recombinant protein increases intracellular Ca^{2+} ($[Ca^{2+}]_i$) signaling in several cell types including adipocytes and stimulates fatty acid and triglyceride synthesis in adipocytes at least in part via a Ca^{2+} -dependent mechanism. Agouti and insulin act in an additive manner to increase fatty acid synthesis in adipocytes. Interestingly, agouti is expressed in human adipose tissue and pancreatic islets where it increases intracellular calcium and insulin secretion.

L5 ANSWER 7 OF 27 BIOSIS COPYRIGHT 1998 BIOSIS DUPLICATE 2
 ACCESSION NUMBER: 98:393792 BIOSIS
 DOCUMENT NUMBER: 01393792
 TITLE: Effects of a potent **melanocortin** agonist on the diabetic-obese phenotype in yellow mice.
 AUTHOR(S): Zemel M B; Moore J W; Moustaid N; Kim J H; Nichols J S; Blanchard S G; Parks D J; Harris C; Lee F W; Grizzle M; James M; Wilkinson W O
 CORPORATE SOURCE: Zen-Bio Inc., 3200 Chapel Hill, Nelson Blvd., Suite 102, P.O. Box 12503, Research Triangle Park, NC 27709, USA
 SOURCE: International Journal of Obesity 22 (7). 1998. 678-683. ISSN: 0307-0565
 LANGUAGE: English
 AB OBJECTIVE: To test the hypothesis that a **melanocortin** agonist can reverse obesity and insulin resistance in mice overexpressing the agouti protein, EXPERIMENTAL MODEL: Mice overexpressing the agouti protein either by **transgene** introduction (beta-actin promotor) or by mutation (AY). DESIGN: NDPMSH was tested for pharmacokinetic suitability. NDPMSH at various doses was administered subcutaneously twice a day for 2-3 weeks. MEASUREMENTS: Fur pigmentation, various fatness parameters (core temperature, fat pad weight and body weight), blood glucose and hormones, fatty acid synthase measurement. RESULTS: NDPMSH caused fur pigmentation and core temperature changes, but failed to affect any metabolic parameters in agouti-dependent manner. CONCLUSION: NDPMSH, as a representation **melanocortin** agonist, does not compete with agouti in reversing agouti-dependent metabolic effects. This suggests that 1) agouti works via a receptor other than a **melanocortin** receptor to mediate its metabolic effects, 2) agouti-dependent metabolic effects are mediated through **melanocortin** receptors but not via antagonism of these receptors, or 3) NDPMSH is pharmacodynamically an inappropriate molecule for these types of studies.

L5 ANSWER 8 OF 27 BIOSIS COPYRIGHT 1998 BIOSIS DUPLICATE 3
 ACCESSION NUMBER: 98:386925 BIOSIS
 DOCUMENT NUMBER: 01386925
 TITLE: Evidence that orexigenic effects of **melanocortin** 4 receptor antagonist HS014 are mediated by neuropeptide Y.
 AUTHOR(S): Kask A; Rago L; Korrovits P; Wikberg J E S; Schioth H B
 CORPORATE SOURCE: Dep. Pharmacol., Univ. Tartu Ulikooli 18, Tartu EE-2400, Estonia

SOURCE: Biochemical and Biophysical Research
Communications 248 (2). 1998. 245-249. ISSN:
0006-291X

LANGUAGE: English

AB Recent studies using **melanocortin-4** receptor (MC4R)

knockout mice and MC4R antagonists have shown that weakening of MC4R-ergic tone increases food intake and causes obesity. In this study, we used the newly discovered selective MC4R antagonist HS014 for increasing food intake in free-feeding rats and evaluated the effects of the NPY Y-1 receptor antagonist 1229U91 and the selective serotonin uptake inhibitor fluoxetine on this increased feeding behavior. 1229U91 (12 nmol, i.c.v.), which alone does not affect food intake, significantly attenuated the orexigenic effects of HS014, whereas 1 and 3 nmol doses of 1229U91 were ineffective. Fluoxetine, which has been shown to inhibit NPY release, inhibited spontaneous food intake and completely blocked the stimulation of food intake by HS014. These data suggest that feeding induced by weakening of the MC4R-ergic tone may be mediated through activation of the NPY-ergic system. This is the first report showing that physiological feeding response evoked by MC4R blockage is influenced by NPY signalling.

L5 ANSWER 9 OF 27 DISSABS COPYRIGHT 1998 UMI Company

ACCESSION NUMBER: 97:59645 DISSABS Order Number: AAR9728495

TITLE: EXTINCTION AND TRANSACTIVATION OF MELANOCYTE-SPECIFIC
GENE EXPRESSION IN MICROCELL HYBRIDS (MICROPHTHALMIA)

AUTHOR: POWERS, THOMAS PATRICK [PH.D.]

CORPORATE SOURCE: UNIVERSITY OF ILLINOIS AT CHICAGO, HEALTH SCIENCES
CENTER (0806)

SOURCE: Dissertation Abstracts International, (1997) Vol. 58,
No. 4B, p. 1706. Order No.: AAR9728495. 184 pages.

DOCUMENT TYPE: Dissertation

FILE SEGMENT: DAI

LANGUAGE: English

AB Pigmentation and regulation of melanocyte-specific gene expression was examined in whole-cell and microcell hybrids between mouse fibroblasts and pigmented Syrian hamster melanoma cells. Extinction of pigmentation was observed in whole-cell hybrids and in microcell hybrids containing very little fibroblast chromosomal material, suggesting that a specific fibroblast extinguisher locus may regulate extinction of pigmentation.

Expression of tyrosinase, TRP-1 (tyrosinase-related protein-1), TRP-2 (tyrosinase-related protein-2), microphthalmia, and MC1R (**melanocortin-1** receptor) genes was analyzed in whole-cell and microcell hybrids, using reverse transcription combined with the polymerase chain reaction. All five cDNAs were undetectable in unpigmented whole-cell hybrids, demonstrating extinction of multiple melanocyte-specific genes in hybrid cells. Similar analyses in microcell hybrids suggested that tyrosinase, TRP-1, and microphthalmia genes were extinguished coordinately while the TRP-2 and MC1R genes were extinguished individually by distinct mechanisms.

A microphthalmia **transgene** was expressed in unpigmented microcell hybrids and pigmentation and melanocyte-specific gene expression were assayed. While expression of microphthalmia did not result in pigmentation or expression of the tyrosinase and TRP-1 genes, it did result in activation of TRP-2 gene expression. These results demonstrate a differential response of melanocyte-specific genes to microphthalmia in hybrid cells. Furthermore, the results imply that extinction of the TRP-2 gene requires two separate fibroblast extinguishers.

Finally, experiments were carried out to determine if melanocyte-specific genes carried on mouse and human fibroblast chromosomes were transactivated in pigmented microcell hybrids. Using the polymerase chain reaction with species-specific PCR primers to detect fibroblast cDNAs, it was shown that mouse

fibroblast tyrosinase, TRP-1, microphthalmia, MC1R, and silver genes were transactivated in a pigmented microcell hybrid. Transactivation of the human tyrosinase gene from fibroblast cells also was demonstrated in pigmented microcell hybrids. These studies demonstrated that melanocyte-specific genes from fibroblast cells could be transactivated when transferred into pigmented melanoma cells.

L5 ANSWER 10 OF 27 COPYRIGHT 1998 IAC

ACCESSION NUMBER: 97:347659 NLDB
TITLE: CHANGE IN MOUSE HAIR COLOR ANOTHER CLUE TO WEIGHT GAIN
SOURCE: BIOWORLD Today, (7 Oct 1997) Vol. 8, No. 194.
PUBLISHER: American Health Consultants Inc.
DOCUMENT TYPE: Newsletter
LANGUAGE: English
WORD COUNT: 991

L5 ANSWER 11 OF 27 COPYRIGHT 1998 IAC

ACCESSION NUMBER: 97:13015 NLDB
TITLE: MILLENNIUM FINDS OBESITY CIRCUIT IN BRAIN INDEPENDENT OF LEPTIN; DRUG DISCOVERY NEXT
SOURCE: BIOWORLD Today, (10 Jan 1997) Vol. 8, No. 7.
PUBLISHER: American Health Consultants
DOCUMENT TYPE: Newsletter
LANGUAGE: English
WORD COUNT: 897

L5 ANSWER 12 OF 27 COPYRIGHT 1998 IAC

ACCESSION NUMBER: 97:385287 NLDB
TITLE: P&G Supports Obesity Research
SOURCE: Applied Genetics News, (1 Nov 1997) Vol. 18, No. 4. ISSN: 0271-7107.
PUBLISHER: Business Communications Company, Inc
DOCUMENT TYPE: Newsletter
LANGUAGE: English
WORD COUNT: 430

L5 ANSWER 13 OF 27 COPYRIGHT 1998 PJB

ACCESSION NUMBER: 97:2856 PHIN
DOCUMENT NUMBER: S00523368
DATA ENTRY DATE: 28 Jan 1997
TITLE: Cerebrus finds satiety mechanism
SOURCE: Scrip (1997) No. 2201 p21
DOCUMENT TYPE: Newsletter
FILE SEGMENT: FULL

L5 ANSWER 14 OF 27 PNI COPYRIGHT 1998 UMI Company

ACCESSION NUMBER: 97:35369 PNI
DOCUMENT NUMBER: 97-35369
TITLE: P&G supports obesity research
SOURCE: Applied Genetics News, (971100) Vol. 18, No. 4, pp. 5-6.
CODEN: AGNEEN; ISSN: 0271-7107.
DOCUMENT TYPE: Newsletter
LANGUAGE: English

L5 ANSWER 15 OF 27 CANCERLIT

DUPLICATE 4

ACCESSION NUMBER: 97426593 CANCERLIT
DOCUMENT NUMBER: 97426593
TITLE: The role of the agouti gene in the yellow obese

syndrome.

AUTHOR: Miltenberger R J; Mynatt R L; Wilkinson J E; Woychik R P

CORPORATE SOURCE: Mammalian Genetics and Development Section, Oak Ridge National Laboratory, Oak Ridge, TN 37831, USA.

SOURCE: JOURNAL OF NUTRITION, (1997). Vol. 127, No. 9, pp. 1902S-1907S.
Journal code: JEV. ISSN: 0022-3166.

DOCUMENT TYPE: Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)

FILE SEGMENT: MEDL; L; Priority Journals

LANGUAGE: English

OTHER SOURCE: MEDLINE 97426593

ENTRY MONTH: 199711

AB The yellow obese syndrome in mice encompasses many pleiotropic effects including yellow fur, maturity-onset obesity, hyperinsulinemia, insulin resistance, hyperglycemia, increased skeletal length and lean body mass, and increased susceptibility to neoplasia. The molecular basis of this syndrome is beginning to be unraveled and may have implications for human obesity and diabetes. Normally, the agouti gene is expressed during the hair-growth cycle in the neonatal skin where it functions as a paracrine regulator of pigmentation. The secreted agouti protein antagonizes the binding of the alpha-melanocyte-stimulating hormone to its receptor (**melanocortin 1** receptor) on the surface of hair bulb melanocytes, causing alterations in intracellular cAMP levels. Widespread, ectopic expression of the mouse agouti gene is central to the yellow obese phenotype, as demonstrated by the molecular cloning of several dominant agouti mutations and the ubiquitous expression of the wild-type agouti gene in **transgenic** mice. Recent experiments have revealed that the hypothalamus and adipose tissue are biologically active target sites for agouti in the yellow obese mutant lines. (81 Refs)

L5 ANSWER 16 OF 27 CAPLUS COPYRIGHT 1998 ACS

ACCESSION NUMBER: 1997:595630 CAPLUS

DOCUMENT NUMBER: 127:276141

TITLE: The role of the agouti gene in the yellow obese syndrome

AUTHOR(S): Miltenberger, Rosalynn J.; Mynatt, Randall L.; Wilkinson, J. Erby; Woychik, Richard P.

CORPORATE SOURCE: Mammalian Genetics and Development Section, Oak Ridge National Laboratory, Oak Ridge, TN, 37831, USA

SOURCE: J. Nutr. (1997), 127(9), 1902S-1907S
CODEN: JONUAI; ISSN: 0022-3166

PUBLISHER: American Society for Nutritional Sciences

DOCUMENT TYPE: Journal; General Review

LANGUAGE: English

AB A review with 81 refs. The yellow obese syndrome in mice encompasses many pleiotropic effects including yellow fur, maturity-onset obesity, hyperinsulinemia, insulin resistance, hyperglycemia, increased skeletal length and lean body mass, and increased susceptibility to neoplasia. The mol. basis of this syndrome is beginning to be unraveled and may have implications for human obesity and diabetes. Normally, the agouti gene is expressed during the hair-growth cycle in the neonatal skin where it functions as a paracrine regulator of pigmentation. The secreted agouti protein antagonizes the binding of the .alpha.-MSH to its receptor (**melanocortin 1** receptor) on the surface of hair bulb melanocytes, causing alterations in intracellular cAMP levels. Widespread ectopic expression of the mouse agouti gene is central to the yellow obese phenotype, as demonstrated by the mol. cloning of several dominant agouti mutations and the ubiquitous expression of

the wild-type agouti gene in **transgenic** mice. Recent expts. have revealed that the hypothalamus and adipose tissue are biol. active target sites for agouti in the yellow obese mutant lines.

L5 ANSWER 17 OF 27 BIOSIS COPYRIGHT 1998 BIOSIS DUPLICATE 5
ACCESSION NUMBER: 97:487558 BIOSIS
DOCUMENT NUMBER: 99786761
TITLE: Genetics of human obesity: Research directions.
AUTHOR(S): Bray G; Bouchard C
CORPORATE SOURCE: Pennington Biomed. Res. Cent., Baton Rouge, LA
70808-4124, USA
SOURCE: FASEB Journal 11 (12). 1997. 937-945. ISSN:
0892-6638
LANGUAGE: English

AB Rapid strides in understanding the physiology controlling energy or nutrient intake and energy expenditure have complemented the search for the genetic basis of obesity. Several single gene defects are known that produce obesity in animals. All of these have been cloned within the past 4 years, providing a rich new base for understanding obesity. Since obesity is likely to be "multifactorial," a number of laboratories have used the quantitative trait locus (QTL) technique of genome scanning to identify candidate genomic regions and, eventually, genes that may influence body weight and body fat. So far, 18 QTLs have been identified in association with crossbreeding strains of mice or rats with variable susceptibility to obesity. A number of mendelian disorders are known to exist in humans, but no specific genes have yet been identified for them. The potential for inserting new genetic material into mammals has produced numerous **transgenic** mice with increased or decreased quantities of body fat. These models will provide a continuing source of new insights into obesity. Several areas in the human genome have been linked to the development of obesity. Among the candidate genes with evidence of linkage to body fat are TNF-alpha, adenosine deaminase, and **melanocortin**-3 receptor. The new insights described above have invigorated the pharmaceutical industry to increase their efforts for new drug development aimed at the growing problem of obesity.

L5 ANSWER 18 OF 27 MEDLINE
ACCESSION NUMBER: 1998074799 MEDLINE
DOCUMENT NUMBER: 98074799
TITLE: Exocrine gland dysfunction in MC5-R-deficient mice: evidence for coordinated regulation of exocrine gland function by **melanocortin** peptides.
AUTHOR: Chen W; Kelly M A; Opitz-Araya X; Thomas R E; Low M J; Cone R D
CORPORATE SOURCE: Vollum Institute, Oregon Health Sciences University, Portland 97201, USA.
CONTRACT NUMBER: AR42415 (NIAMS)
HD30236 (NICHD)
SOURCE: CELL, (1997 Dec 12) 91 (6) 789-98.
Journal code: CQ4. ISSN: 0092-8674.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199803
ENTRY WEEK: 19980303

AB The effects of pituitary-derived **melanocortin** peptides are primarily attributed to ACTH-mediated adrenocortical glucocorticoid production. Identification of a widely distributed receptor for ACTH/MSH peptides, the **melanocortin**-5 receptor (MC5-R), suggested non-steroidally mediated systemic effects of these peptides. Targeted disruption of the MC5-R produced mice with a

severe defect in water repulsion and thermoregulation due to decreased production of sebaceous lipids. High levels of MC5-R was found in multiple exocrine tissues, including Harderian, preputial, lacrimal, and sebaceous glands, and was also shown to be required for production and stress-regulated synthesis of porphyrins by the Harderian gland and ACTH/MSH-regulated protein secretion by the lacrimal gland. These data show a requirement for the MC5-R in multiple exocrine glands for the production of numerous products, indicative of a coordinated system for regulation of exocrine gland function by **melanocortin** peptides.

L5 ANSWER 19 OF 27 BIOSIS COPYRIGHT 1998 BIOSIS DUPLICATE 6
ACCESSION NUMBER: 97:254483 BIOSIS
DOCUMENT NUMBER: 99553686
TITLE: Induction of neuropeptide Y gene expression in the dorsal medial hypothalamic nucleus in two models of the agouti obesity syndrome.
AUTHOR(S): Kesterson R A; Huszar D; Lynch C A; Simerly R B; Cone R D
CORPORATE SOURCE: Vollum Inst. Advanced Biomed. Research, Oregon Health Sci. Univ., 3181 Southwest Sam Jackson Park Road, Portland, OR 97201-3098, USA
SOURCE: Molecular Endocrinology 11 (5). 1997. 630-637. ISSN: 0888-8809
LANGUAGE: English

AB Dominant mutations at the agouti locus induce several phenotypic changes in the mouse including yellow pigmentation (phaeomelanization) of the coat and adult-onset obesity. Nonpigmentary phenotypic changes associated with the agouti locus are due to ectopic expression of the agouti-signaling protein (ASP), and the pheomelanizing effects on coat color are due to ASP antagonism of alpha-MSH binding to the melanocyte MC1 receptor. Recently it has been demonstrated that pharmacological antagonism of hypothalamic **melanocortin** receptors or genetic deletion of the **melanocortin** 4 receptor (MC4-R) recapitulates aspects of the agouti obesity syndrome, thus establishing that chronic disruption of central melanocortinerbic signaling is the cause of agouti-induced obesity. To learn more about potential downstream effectors involved in these melanocortinerbic obesity syndromes, we have examined expression of the orexigenic peptides galanin and neuropeptide Y (NPY), as well as the anorexigenic POMC in lethal yellow (A-y), MC4-R **knockout** (MC4-RKO), and leptin-deficient (ob/ob) mice. No significant changes in galanin or POMC gene expression were seen in any of the obese models. In situ hybridizations using an antisense NPY probe demonstrated that in obese A-y mice, arcuate nucleus NPY mRNA levels were equivalent to that of their C57BL/6J littermates. However, NPY was expressed at high levels in a new site, the dorsal medial hypothalamic nucleus (DMH). Expression of NPY in the DMH was also seen in obese MC4-RKO homozygous (-/-) mice, but not in lean heterozygous (+/-) or wild type (+/+) control mice. This identifies the DMH as a brain region that is functionally altered by the disruption of melanocortinerbic signaling and suggests that this nucleus, possibly via elevated NPY expression, may have an etiological role in the melanocortinerbic obesity syndrome.

L5 ANSWER 20 OF 27 JICST-EPlus COPYRIGHT 1998 JST
ACCESSION NUMBER: 980224059 JICST-EPlus
TITLE: **Knockout** Mouse Data Book.
Melanocortin-4 receptor(MC4R).
AUTHOR: MORI YASUMICHI
KADOWAKI TAKASHI
CORPORATE SOURCE: Asahi Life Found., Inst. for Diabetes Care and Res. Univ. of Tokyo
SOURCE: Mol Med, (1997) vol. 34, no. Dec rinji zokango, pp. 313-314. Journal Code: Z0625A

PUB. COUNTRY: Japan
LANGUAGE: Japanese
STATUS: New

CODEN: MOLMEL; ISSN: 0918-6557

L5 ANSWER 21 OF 27 CAPLUS COPYRIGHT 1998 ACS DUPLICATE 7
ACCESSION NUMBER: 1997:700359 CAPLUS
DOCUMENT NUMBER: 128:12357
TITLE: Overexpression of Agrt leads to obesity in
transgenic mice
AUTHOR(S): Graham, Melissa; Shutter, John R.; Sarmiento,
Ulla; Sarosi, Ildiko; Stark, Kevin L.
CORPORATE SOURCE: Dep. Molecular Genetics, Thousand Oaks, CA,
91320, USA
SOURCE: Nat. Genet. (1997), 17(3), 273-274
CODEN: NGENEC; ISSN: 1061-4036
PUBLISHER: Nature America
DOCUMENT TYPE: Journal
LANGUAGE: English

AB **Transgenic** mice overexpressing the Agrt gene were produced using mouse Agrt cDNA under the control of the human .beta.-actin promoter. In addn. to overexpression of Agrt in brain tissue, Agrt mRNA was detected in the skin, skeletal muscle, liver and white adipose tissue from the **transgenic** but not nontransgenic mice. Overexpression of Agrt recapitulated many unique features of the obese yellow agouti and **melanocortin 4** receptor-deficient mice: obesity, increased body length, hyperinsulinemia, late-onset hyperglycemia, pancreatic islet hyperplasia and hypertrophy, and lack of elevated corticosterone.

L5 ANSWER 22 OF 27 CAPLUS COPYRIGHT 1998 ACS
ACCESSION NUMBER: 1997:718678 CAPLUS
DOCUMENT NUMBER: 128:21146
TITLE: Obesity and the adipocyte. Role of the agouti gene in obesity
AUTHOR(S): Michaud, E. J.; Mynatt, R. L.; Miltenberger, R. J.; Klebig, M. L.; Wilkinson, J. E.; Zemel, M. B.; Wilkison, W. O.; Woychik, R. P.
CORPORATE SOURCE: Life Sci. Div., Oak Ridge Natl. Lab., Oak Ridge, TN, 37831, USA
SOURCE: J. Endocrinol. (1997), 155(2), 207-209
CODEN: JOENAK; ISSN: 0022-0795
PUBLISHER: Journal of Endocrinology
DOCUMENT TYPE: Journal; General Review
LANGUAGE: English

AB A review, with .apprx.40 refs., suggesting that the wild-type agouti protein acts on both the CNS and tissues in the periphery to induce the obesity syndrome. In the CNS agouti may antagonize neural **melanocortin** receptor(s), resulting in obesity, hyperphagia, and hyperinsulinemia, as obsd. in MC4-R **knockout** mice. In the periphery, agouti expression in adipose tissue, coupled with insulin treatment, results in significant wt. gains in mice. Given that hyperphagia appears to be an important aspect of the agouti-induced obesity syndrome, it is noteworthy that pancreatic beta-cell hyperplasia precedes obesity in mutant agouti mice. In addn., increases in [Ca2+]i in beta cells stimulate insulin release. Therefore, it is possible that ectopic expression of the agouti gene in the pancreas may act directly on the beta cells to trigger hyperplasia.

L5 ANSWER 23 OF 27 BIOSIS COPYRIGHT 1998 BIOSIS DUPLICATE 8
ACCESSION NUMBER: 97:505006 BIOSIS
DOCUMENT NUMBER: 99804209
TITLE: Antagonism of central **melanocortin** receptors in vitro and in vivo by Agouti-related

protein.
AUTHOR(S): Ollmann M M; Wilson B D; Yang Y-K; Kerns J A; Chen Y; Gantz I; Barsh G S
CORPORATE SOURCE: Beckman Cent. B271A, Stanford Univ. Sch. Med.,
Stanford, CA 94305-5323, USA
SOURCE: Science (Washington D C) 278 (5335). 1997.
135-138. ISSN: 0036-8075
LANGUAGE: English

AB Expression of Agouti protein is normally limited to the skin where it affects pigmentation, but ubiquitous expression causes obesity. An expressed sequence tag was identified that encodes Agouti-related protein, whose RNA is normally expressed in the hypothalamus and whose levels were increased eightfold in ob/ob mice. Recombinant Agouti-related protein was a potent, selective antagonist of Mc3r and Mc4r, **melanocortin** receptor subtypes implicated in weight regulation. Ubiquitous expression of human AGRP complementary DNA in **transgenic** mice caused obesity without altering pigmentation. Thus, Agouti-related protein is a neuropeptide implicated in the normal control of body weight downstream of leptin signaling.

L5 ANSWER 24 OF 27 MEDLINE

ACCESSION NUMBER: 97148695 MEDLINE
DOCUMENT NUMBER: 97148695
TITLE: Targeted disruption of the **melanocortin-4** receptor results in obesity in mice.
AUTHOR: Huszar D; Lynch C A; Fairchild-Huntress V; Dunmore J H; Fang Q; Berkemeier L R; Gu W; Kesterson R A; Boston B A; Cone R D; Smith F J; Campfield L A; Burn P; Lee F
CORPORATE SOURCE: Millennium Pharmaceuticals, Inc., Cambridge, Massachusetts 02139, USA.
SOURCE: CELL, (1997 Jan 10) 88 (1) 131-41.
Journal code: CQ4. ISSN: 0092-8674.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
LANGUAGE: English
FILE SEGMENT: Priority Journals; Cancer Journals
ENTRY MONTH: 199704
ENTRY WEEK: 19970403

AB The **melanocortin-4** receptor (MC4-R) is a G protein-coupled, seven-transmembrane receptor expressed in the brain. Inactivation of this receptor by gene targeting results in mice that develop a maturity onset obesity syndrome associated with hyperphagia, hyperinsulinemia, and hyperglycemia. This syndrome recapitulates several of the characteristic features of the agouti obesity syndrome, which results from ectopic expression of agouti protein, a pigmentation factor normally expressed in the skin. Our data identify a novel signaling pathway in the mouse for body weight regulation and support a model in which the primary mechanism by which agouti induces obesity is chronic antagonism of the MC4-R.

L5 ANSWER 25 OF 27 MEDLINE

ACCESSION NUMBER: 97314693 MEDLINE
DOCUMENT NUMBER: 97314693
TITLE: Neuropeptides responding to leptin.
AUTHOR: Wolf G
CORPORATE SOURCE: Department of Nutritional Sciences, University of California, Berkeley 94720-3104, USA.
SOURCE: NUTRITION REVIEWS, (1997 Mar) 55 (3) 85-8. Ref: 20
Journal code: OAY. ISSN: 0029-6643.
PUB. COUNTRY: United States
Journal; Article; (JOURNAL ARTICLE)
General Review; (REVIEW)
(REVIEW, TUTORIAL)
LANGUAGE: English

ENTRY MONTH: 199708
ENTRY WEEK: 19970803

AB Leptin, the circulating protein that inhibits food intake and energy expenditure, was thought to function through inhibition of the hypothalamic neuropeptide Y (NPY), a stimulator of food intake. However, mouse mutants lacking NPY are normal, suggesting that alternative neuromodulators of food intake must exist. Recently, **melanocortin**, a neuropeptide acting on the hypothalamic receptor melanocortin4-R, was discovered in mice, controlling energy regulation. This receptor is antagonized by the "agouti" protein in the mutant obese agouti mouse.

L5 ANSWER 26 OF 27 BIOSIS COPYRIGHT 1998 BIOSIS DUPLICATE 9
ACCESSION NUMBER: 96:461724 BIOSIS
DOCUMENT NUMBER: 99184080
TITLE: Coupled site-directed mutagenesis-
transgenesis identifies important
functional domains of the mouse agouti protein.
AUTHOR(S): Perry W L; Nakamura T; Swing D A; Secrest L;
Eagleson B; Hustad C M; Copeland N G; Jenkins N A
CORPORATE SOURCE: Mammalian Genetics Lab., ABL-Basic Research
Program, NCI-Frederick Cancer Research Development
Center, P.O. Box B, Frederick, MD 21702, USA
SOURCE: Genetics 144 (1). 1996. 255-264. ISSN: 0016-6731
LANGUAGE: English

AB The agouti locus encodes a novel paracrine signaling molecule containing a signal sequence, an N-linked glycosylation site, a central lysine-rich basic domain, and a C-terminal tail containing 10 cysteine (Cys) residues capable of forming five disulfide bonds. When overexpressed, agouti causes a number of pleiotropic effects including yellow coat and adult-onset obesity. Numerous studies suggest that agouti causes yellow coat color by antagonizing the binding of alpha-melanocyte-stimulating hormone (alpha-MSH) to the alpha-MSH-(**melanocortin**-1) receptor. With the goal of identifying functional domains of agouti important for its diverse biological activities, we have generated 14 agouti mutations by in vitro site-directed mutagenesis and analyzed these mutations in **transgenic** mice for their effects on coat color and obesity. These studies demonstrate that the signal sequence, the N-linked glycosylation site, and the C-terminal Cys residues are important for full biological activity, while at least a portion of the lysine-rich basic domain is dispensable for normal function. They also show that the same functional domains of agouti important in coat color determination are important for inducing obesity, consistent with the hypothesis that agouti induces obesity by antagonizing **melanocortin** binding to other **melanocortin** receptors.

L5 ANSWER 27 OF 27 BIOSIS COPYRIGHT 1998 BIOSIS
ACCESSION NUMBER: 94:60940 BIOSIS
DOCUMENT NUMBER: 97073940
TITLE: Rat and mouse proopio-**melanocortin** gene
sequences target tissue-specific expression to the
pituitary gland but not to the hypothalamus of
transgenic mice.
AUTHOR(S): Rubinstein M; Mortrud M; Liu B; Low M J
CORPORATE SOURCE: Vollum Inst. Adv. Biomed. Res., Oreg. Health Sci.
Univ. L-474, 3181 Sam Jackson Park Rd., Portland,
OR 97201, USA
SOURCE: Neuroendocrinology 58 (4). 1993. 373-380. ISSN:
0028-3835
LANGUAGE: English

AB The proopiomelanocortin (POMC) gene is expressed predominantly in corticotrophs of the pituitary anterior lobe, melanotrophs of the intermediate lobe and neurons of the arcuate nucleus of the

hypothalamus. The different ontogeny of POMC mRNA as well as the complicated hormonal regulation of POMC gene expression in the three different cell types suggests a concerted interaction between several cis-acting elements in the POMC gene and transcription factors located in each of the three cell types. To investigate cell-specific elements in the POMC gene we tested two different constructs in **transgenic** mice. The construct -4000rPOMCLacZ, carrying 4 kb of the rat POMC promoter fused to the Escherichia coli beta-galactosidase gene, showed appropriate expression in melanotrophs in 50% of the mice analyzed. beta-Galactosidase activity was less evident in corticotrophs under basal environmental conditions. In brain, 7 out of 15 independently derived **transgenic** founders had ectopic expression of the **transgene** in different areas; however, none of the animals analyzed expressed beta-galactosidase in neurons of the arcuate nucleus. The construct HAL*, a 'tagged' 10.2-kb mouse genomic fragment, was more efficiently targeted to the pituitary. Using in situ hybridization, we detected uniform expression of HAL* in melanotrophs in 100% of the 6 pedigrees analyzed and **transgenic** mRNA levels paralleled those of the endogenous POMC mRNA. In corticotrophs, basal expression was low but after adrenalectomy HAL* mRNA levels were comparable to those of POMC. None of the 6 pedigrees had appropriate expression of HAL* in the brain; however, 2 lines had ectopic expression in the dentate gyrus of the hippocampus. These data, together with previously reported studies, suggest that accurate neuronal expression of the POMC gene requires DNA elements in addition to the sequences that are sufficient for expression in pituitary melanotrophs and corticotrophs. The consistently lower level of **transgene** expression in corticotrophs compared to melanotrophs under basal conditions indicates that these two pituitary cell types may also differ in their requirements for POMC gene regulatory elements.

L9 ANSWER 2 OF 18 CAPLUS COPYRIGHT 1998 ACS

ACCESSION NUMBER: 1998:398429 CAPLUS

DOCUMENT NUMBER: 129:64011

TITLE: **Syndecan** enhancer element and its use
for targeting gene expressionINVENTOR(S): Jalkanen, Markku; Jaakkola, Panu; Vihinen,
TapaniPATENT ASSIGNEE(S): Oy Biotie Therapies, Ltd., Finland; Jalkanen,
Markku; Jaakkola, Panu; Vihinen, TapaniSOURCE: PCT Int. Appl., 57 pp.
CODEN: PIXXD2

	NUMBER	DATE
	-----	-----
PATENT INFORMATION:	WO 9824921 A1	19980611
DESIGNATED STATES:	W: AL, AM, AU, AZ, BA, BG, BR, BY, CA, CN, CZ, EE, GE, HU, ID, IL, IS, JP, KG, KR, KZ, LT, LV, MD, MK, MX, NO, NZ, PL, RO, RU, SG, SI, SK, TJ, TM, TR, UA, US, UZ, YU, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM RW: AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE	
APPLICATION INFORMATION:	WO 97-FI748	19971202
PRIORITY APPLN. INFO.:	US 96-760534	19961202
DOCUMENT TYPE:	Patent	
LANGUAGE:	English	

AB A **syndecan** enhancer element, novel proteins that activate the enhancer element, non-human **transgenic** animals comprising this enhancer element linked to a structural gene, and the use of this enhancer element to regulate the expression of **syndecan** and other genes are also provided. Regulatory elements assocd. with the **syndecan**-1 gene enhancer are the fibroblast growth factor-inducible response element (FiRE). Expression vectors are also prepd. which contain the SV40 promoter or the thymidine kinase gene promoter. The enhancer element can also be used to target expression of a gene to wound sites. The enhancer element and targeted gene expression are exemplified in **transgenic mouse** models.

L9 ANSWER 3 OF 18 CAPLUS COPYRIGHT 1998 ACS
 ACCESSION NUMBER: 1998:324878 CAPLUS
 DOCUMENT NUMBER: 128:318018
 TITLE: Construction of **transgenic mice** expressing a **syndecan 1** gene in regions of hypothalamus for wt. regulation and therapy
 INVENTOR(S): Bernfield, Merton; Reizes, Ofer
 PATENT ASSIGNEE(S): Children's Medical Center Corp., USA
 SOURCE: PCT Int. Appl., 61 pp.
 CODEN: PIXXD2

	NUMBER	DATE
	-----	-----
PATENT INFORMATION:	WO 9820121 A1	19980514
DESIGNATED STATES:	W: AL, AU, BA, BB, BG, BR, CA, CN, CU, CZ, EE, GE, HU, IL, IS, JP, KP, KR, LC, LK, LR, LT, LV, MG, MK, MN, MW, MX, NO, NZ, PL, RO, SG, SI, SK, SL, TR, TT, UA, UZ, VN, YU, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM	
	RW: AT, BE, BF, BJ, CF, CG, CH, CI, CM, DE, DK, ES, FI, FR, GA, GB, GR, IE, IT, LU, MC, ML, MR, NE, NL, PT, SE, SN, TD, TG	
APPLICATION INFORMATION:	WO 97-US20003	19971106
PRIORITY APPLN. INFO.:	US 96-30758	19961106
DOCUMENT TYPE:	Patent	
LANGUAGE:	English	

AB Lines of **transgenic mice** have been developed which preferentially express a **syndecan** in the regions of the hypothalamus which are known to be important in wt. control. The animals were made using a construct including a cytomegalovirus promoter and the 3' untranslated region, including the polyadenylation site, of the bovine growth hormone gene, as well as cDNA encoding **syndecan-1**. The **mice** express the **syndecan-1 transgene** in many tissues, with the expression in the brain occurring preferentially in their hypothalamus. The **mice** are characterized by elevated levels of circulating **syndecan-1** ectodomain and exhibit enormous wt. gain after reaching sexual maturity. **Transgenic** animals in which stop codons have been inserted into the construct so that the **syndecan** is not expressed do not exhibit the same enormous wt. gain. The animals have a relatively normal distribution of fat, are completely healthy and heterozygotes reproduce, and show other indicators assocd. with obesity in humans. The **mice** are useful in understanding the factors involved in wt. regulation and in designing and screening for drugs which are involved in wt. regulation and that can either enhance or reduce appetite and activity. Wasting disorders which can be examd. in these **mice** include idiopathic obesity, anorexia nervosa, and cachexia due to cancer, cancer chemotherapy, chronic inflammatory disease, rheumatoid and collagen diseases, and chronic infections.

L9 ANSWER 5 OF 18 TOXLIT
ACCESSION NUMBER: 1998:77126 TOXLIT
DOCUMENT NUMBER: CA-128-318018P
TITLE: Construction of **transgenic mice**
expressing a **syndecan** 1 gene in regions of
hypothalamus for wt. regulation and therapy.
AUTHOR: Bernfield M; Reizes O
SOURCE: (1998). PCT Int. Appl. PATENT NO. 9820121 05/14/1998
(Children's Medical Center Corp.).
CODEN: PIXXD2.
PUB. COUNTRY: UNITED STATES
DOCUMENT TYPE: Patent
FILE SEGMENT: CA
LANGUAGE: English
OTHER SOURCE: CA 128:318018
ENTRY MONTH: 199806

AB Lines of **transgenic mice** have been developed
which preferentially express a **syndecan** in the regions of
the hypothalamus which are known to be important in wt. control. The
animals were made using a construct including a cytomegalovirus
promoter and the 3' untranslated region, including the
polyadenylation site, of the bovine growth hormone gene, as well as
cDNA encoding **syndecan**-1. The **mice** express the
syndecan-1 **transgene** in many tissues, with the
expression in the brain occurring preferentially in their
hypothalamus. The **mice** are characterized by elevated
levels of circulating **syndecan**-1 ectodomain and exhibit
enormous wt. gain after reaching sexual maturity. **Transgenic**
animals in which stop codons have been inserted into the construct
so that the **syndecan** is not expressed do not exhibit the
same enormous wt. gain. The animals have a relatively normal
distribution of fat, are completely healthy and heterozygotes
reproduce, and show other indicators assocd. with obesity in humans.
The **mice** are useful in understanding the factors involved
in wt. regulation and in designing and screening for drugs which are
involved in wt. regulation and that can either enhance or reduce
appetite and activity. Wasting disorders which can be examd. in
these **mice** include idiopathic obesity, anorexia nervosa,
and cachexia due to cancer, cancer chemotherapy, chronic
inflammatory disease, rheumatoid and collagen diseases, and chronic
infections.

L9 ANSWER 9 OF 18 BIOSIS COPYRIGHT 1998 BIOSIS DUPLICATE 3
ACCESSION NUMBER: 98:391437 BIOSIS
DOCUMENT NUMBER: 01391437
TITLE: Screening downstream genes of a homeobox gene by
differential display using a **knockout**
mouse.
AUTHOR(S): Nishizawa K; Satokata I; Kawano Y; Uchiyama M
CORPORATE SOURCE: Dep. Pediatrics, Niigata Univ. Sch. Med., 1-754
Asahimachi-dori, Niigata 951-8510, Japan
SOURCE: Acta Medica et Biologica 46 (2). 1998. 63-70.
ISSN: 0567-7734
LANGUAGE: English

AB The products of homeobox genes are DNA-binding transcription factors. However, little is known about downstream genes whose activities are regulated directly or indirectly by the homeobox genes. In the current study, we tested a differential display (DD) method using the tissue of a **knockout mouse** in order to identify the downstream genes of a homeobox gene *Msx1* systematically. Our previous in situ hybridization analysis of a *Msx1* deficient **mouse** showed that *Msx1* induced by epithelial bone morphogenetic protein 4 (BMP4) and fibroblast growth factors (FGFs) induces *Bmp4*, the HMG box gene *Lef1*, and the heparan sulfate proteoglycan **syndecan-1** in the tooth mesenchyme. Although it is a very powerful approach for identifying downstream genes of a homeobox gene to test whether the candidate gene's expression is affected in the **knockout mouse**, this approach is not directly applicable to the identification of unknown genes downstream of *Msx1*. In the current study, we performed DD using total RNA from E14.5 *Msx1* mutant mandibles and were able to obtain four novel downstream genes of *Msx1* from 20 cDNA clones verified by Northern blot hybridization and semiquantitative RT-PCR. Despite several problems inherent to this method, we concluded that DD analysis using the tissue of a **knockout mouse** is a useful systematic approach for the identification of downstream genes of a homeobox gene.

L9 ANSWER 11 OF 18 BIOSIS COPYRIGHT 1998 BIOSIS DUPLICATE 4
 ACCESSION NUMBER: 97:249527 BIOSIS
 DOCUMENT NUMBER: 99548730
 TITLE: Use of gene-manipulated models to study the physiology of lipid transport.
 AUTHOR(S): Mortimer B-C; Martins I; Zeng B J; Redgrave T G
 CORPORATE SOURCE: Dep. Physiol., Univ. Western Australia, Nedlands, WA 6907, Australia
 SOURCE: Clinical and Experimental Pharmacology and Physiology 24 (3-4). 1997. 281-285. ISSN: 0305-1870
 LANGUAGE: English

AB 1. In vivo and in vitro gene-manipulated models were used to study the metabolism of chylomicron remnants. **Transgenic mice** expressing human apolipoprotein (Apo) A1 or E4, gene **knockout mice** deficient in ApoE or low density lipoprotein (LDL) receptors and antisense gene inhibition in HepG2 cells were used to evaluate the effect of gene manipulations on the metabolism of chylomicron remnants. 2. **Mice transgenic** for human ApoE4 showed accelerated clearance of chylomicron-like emulsions when animals were fed a low-fat diet. When challenged by a high-fat diet, remnant clearance in ApoE4 **transgenic mice** was delayed, as in normal or non-**transgenic** controls. However, unlike normal nontransgenic controls, in ApoE4 **transgenic mice** high density lipoprotein (HDL)-cholesterol levels remained high after high-fat feeding, which probably protected the animals from the development of atherosclerosis. In contrast, clearance of chylomicron-like lipid emulsions was not affected by the overexpression of human ApoA1 in **transgenic mice**. 3. Gene knock-out **mice** deficient in ApoE or deficient in the LDL receptor were used to show that ApoE and LDL receptors are both essential for the normal, fast catabolism of chylomicron remnants by the liver. 2 In the absence of the LDL receptor, an alternative ApoE-dependent pathway operates to clear chylomicrons from the plasma, with significantly delayed catabolism. 4. Antisense gene inhibition techniques were used to suppress the expression of **syndecan**, a core protein of heparan sulfate proteoglycan, in HepG2 cells. Remnant uptake in cells transfected with the antisense oligodeoxynucleotide complementary to a 20 nucleotide sequence upstream of the initiation site of **syndecan** cDNA markedly reduced the uptake of chylomicron remnant.